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Effects of potato resistant starch intake on insulin sensitivity, related metabolic markers and appetite ratings in men and women at risk for type 2 diabetes: a pilot cross-over randomised controlled trial

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Abstract

Background: The intake of certain types of resistant starch (RS) has been associated in some studies with increased whole-body insulin sensitivity. This randomised, cross-over pilot trial evaluated the effect of consuming cooked, then chilled potatoes, a source of RS, compared to isoenergetic, carbohydrate (CHO)-containing control foods, on insulin sensitivity and related markers.
Methods: Nineteen adults with body mass index 27.0-39.9 kg m^{-2} consumed 300 g day^{-1} RS-enriched potatoes (approximately two potatoes; ~18 g RS) or CHO-based control foods, as part of lunch, evening and snack meals, over a 24-h period. After an overnight fast, insulin sensitivity, CHO metabolism markers, free fatty acids, breath hydrogen levels and appetite were assessed for up to 5 h after the intake of a standard breakfast. The primary endpoint was insulin sensitivity, assessed with the Matsuda index. $P < 0.05$ (one-sided) was considered statistically significant.
Results: Insulin sensitivity was not significantly different between the potato and control conditions. The potato intervention resulted in higher postprandial breath hydrogen ($P = 0.037$), lower postprandial free fatty acid concentrations ($P = 0.039$) and lower fasting plasma glucose ($P = 0.043$) compared to the control condition. Fullness ratings were significantly lower after potato versus control ($P = 0.002$). No other significant effects were observed; however, there was a trend toward lower fasting insulin ($P = 0.077$) in the potato versus the control condition.
Conclusions: The results of this pilot study suggest RS-enriched potatoes may have a favourable impact on carbohydrate metabolism and support the view that additional research in a larger study sample is warranted.

Introduction

Globally, it is estimated that more than 10% of the population will have diabetes by 2030 and, currently, one in two people with diabetes are undiagnosed [(1)]. According to the Centers for Disease Control and Prevention, more than 30 million Americans have diabetes, of which 7.2 million are undiagnosed [(2)]. It has also been estimated that approximately 374 million people around the globe have impaired glucose tolerance and 84 million US adults have prediabetes, placing them at high risk for developing type 2 diabetes (T2D), heart disease and stroke [(1,2)].

Consumption of various types of RS has been reported to enhance whole-body insulin sensitivity, although this finding has not been universal [3-7]. Reduced insulin sensitivity (i.e. insulin resistance) is a major risk factor for the development of T2D, and various interventions that increase insulin sensitivity have been shown to reduce the incidence of new-onset T2D [8]. It has been hypothesised that the mechanism by which resistant starch (RS) may impact insulin resistance is through fermentation of RS by gut microbiota. RS is not fully digested in the small intestine and thus behaves physiologically like a combination of starch and dietary fibre [9]. RS can be fermented to short chain fatty acids (acetate, propionate and butyrate), which are absorbed into the circulation and have various metabolic effects [9-11]. One effect is to inhibit the action of hormone sensitive lipase, thus reducing free fatty acid (FFA) release from the adipocytes [12]. Elevation of plasma FFA levels for several hours (e.g. with an infused lipid emulsion) induces insulin resistance, and reducing FFA levels for several hours (e.g. with the niacin analogue acipimox) enhances insulin sensitivity [13-15].

RS is classified as RS type 1 through RS type 5, depending on the degree of the digestion process that it can resist and the method of formation [10]. Starch can escape digestion in the small intestine as a result of physical inaccessibility within the food matrix (RS type 1) or within starch granules (RS type 2) [16]. Retrograded starch, produced through food manufacturing or preparation, is termed RS type 3, and starch which is chemically modified to a form resistant to digestion ingredient is termed RS type 4. Finally, amylose-lipid complexes comprise RS type 5 [10,16].

Potatoes, especially baked or boiled potatoes that are subsequently chilled, are a dietary source of RS type 3, and therefore may be able to impact insulin resistance through the mechanisms described above [17]. Most studies evaluating RS and insulin sensitivity have administered RS type 2 ingredients at high levels of intake (28 g day^{-1} to 60 g day^{-1}) [4-6,18-20] and/or for longer durations [4,5,18,19]. To our knowledge, only one recent study has evaluated potato RS type 3 at a lower level of intake (10–11 g) for a short time period (<24 h) and assessed glucose and insulin response, but not measure insulin sensitivity [21]. This pilot study was designed to assess effects of short-term consumption of an achievable level of RS intake from a commonly consumed food on insulin sensitivity and other aspects of carbohydrate metabolism.

Based on the results from previous studies conducted by our group, [3,22] the present pilot trial was designed to assess the metabolic effects of consuming 300 g of cooked then chilled potatoes (approximately two whole potatoes) containing ~18 g of RS, over a 24-h period, compared to a control diet matched for calories and macronutrients but with <1 g of RS, on whole-body insulin sensitivity, postprandial insulin, glucose, FFA, appetite ratings and breath hydrogen in overweight or obese adults with an elevated waist circumference. The primary outcome variable was the Matsuda insulin sensitivity index (MISI) calculated from glucose and insulin responses after a standard breakfast meal, and secondary outcome variables were assessments of postprandial glucose, insulin, FFA, breath hydrogen and subjective satiety ratings. It was hypothesised that potato RS consumption would improve insulin sensitivity, increase breath hydrogen (an indication of intestinal fermentation and thus liberation of short-chain fatty acids) and reduce plasma levels of FFAs, particularly in the late-postprandial period after a standard meal. Increased breath hydrogen and reduced FFA would provide the physiological justification for expecting enhanced insulin sensitivity.

Materials and methods

Participants

Adults aged 18-74 years with a body mass index (BMI) of 27.0-39.99 kg m^{-2} and waist circumference >102 cm for men and >89 cm for women were included in the study [23]. The subjects were recruited between September 2018 and January 2019 through the clinic's database of participants who had participated in prior research studies, as well as via social media and referrals from existing participants. Subjects were excluded if they had a fasting capillary glucose of ≥7 mmol L^{-1} and/or a glycated haemoglobin (HbA1C) ≥ 6.5 % (48 mmol mol^{-1}) based on a capillary blood sample, history or presence of atherosclerotic cardiovascular disease, chronic inflammatory disease or clinically important endocrine (e.g. type 1 diabetes or T2D), pulmonary, hepatic, renal, haematological, immunological, dermatological, neurological, psychiatric or biliary disorders, and a history of cancer in the prior 5 years (other than non-melanoma skin cancer). This information was obtained through medical histories provided by the subjects. Individuals were also excluded if they had experienced a change in body weight of ±4.5 kg over the 3 months prior to enrolment. The study was conducted according to Good Clinical Practice Guidelines, the Declaration of Helsinki (2000) [24] and US 21 Code of Federal Regulations and informed consent was obtained from all subjects.

Test foods

Both the potato intervention and the control condition included two low-fibre, RS-free standard breakfast meals a lunch and evening meal and an evening snack. The potato and control conditions were matched for energy

Table 1 Energy, macronutrient, dietary fibre and resistant starch (RS) content of each meal, snack and standardised breakfast meals for the potato-containing foods and carbohydrate (CHO)-based control foods during each treatment condition*

	Potato-containing foods			Isocaloric, CHO-based control foods			Standardised breakfast (both conditions)
Parameter	Lunch & evening meals: potato salad, turkey sandwich with Swiss cheese†	Evening snack: chilled potato gazpacho soup, crackers & juice	Total‡	Lunch & evening meals: noodle salad, turkey sandwich with Swiss cheese†	Evening snack: chilled crouton gazpacho soup, crackers & juice	Total‡	Kellogg's® Corn Pops®, 2% milk
Energy (kJ / kcal)	2172/519	2176/520	8620/2060	2176/520	2172/519	8624/2061	2100/502
Total fat (g)	24	12	69	24	12	69	9
SFA (g)	7.5	0.7	21.6	7.5	0.7	21.6	5.9
Total CHO (g)	51	93	276	47	91	266	81
Dietary fibre (g)	2.6	8.0	13.6	1.9	7.2	11.4	<0.5‡‡
RS** (g)	6.1	6.1	19.2	0.2	0.3	1.6	0.9
FMS†† (g)	40.8	55.7	170.6	44.1	59.8	181.3	33.3
Protein (g)	28	11	86	29	11	88	19

FMS, fully metabolisable starch; SFA, saturated fatty acids.
*Nutrient composition was determined using food label information, when available, and US Department of Agriculture Food Data Central [29].
†Lunch and evening meals were identical.
‡Includes standardised breakfast for both conditions.
**Resistant starch content was estimated based on published tables [17,28,29].
††Estimate based on Total CHO – sugar – dietary fibre – RS.
‡‡Product label states 0 g.

and macronutrients (Table 1). Meals were designed to provide total daily energy similar to the energy values typically consumed, which was confirmed through analysis of 3-day diet records (see Supporting information, Table S1). The potato intervention provided 100 g of Yukon Gold potatoes with the lunch, evening meal and snack meal, for a total of 300 g day^{-1} of potato. Yukon Gold potatoes were chosen as a commonly consumed potato that is semi-starchy and semi-waxy, making it ideal for the recipe preparations in the present study (potato salad and soup). It has been previously shown that the RS3 formation in Yukon Gold potatoes is similar to other varieties of starchy and waxy potatoes [17]. Potatoes were baked in a pre-heated, convection oven at 232 $^{\circ}C$ for 40 min, allowed to cool at room temperature for 10 min, and then chilled at 4 $^{\circ}C$ overnight (15 h) before being sliced, without peeling, and mixed into a potato salad or a chilled gazpacho soup. Baking was chosen instead of boiling to maximise RS formation [17] and chilling overnight has been shown to increase RS in potatoes [25,26]. Test and control foods were prepared in one batch and individual servings were frozen at −20 $^{\circ}C$ until the day of testing to slow further RS development [26]. Each 100 g serving of potato provided ~6 g of RS according to previous published reports on cooked and chilled Yukon Gold potatoes [17], thus providing ~18 g day^{-1} of RS. The control condition provided isocaloric, CHO-based, low-RS lunch and evening meals and an evening snack. In the control lunch and evening meals, egg noodles replaced potatoes in the salad. The control evening snack used toasted, crustless white bread in place of potatoes in the gazpacho. The low-fibre (<1 g), RS-free standard breakfast meal consisted of Kellogg's® Corn Pops® (The Kellogg Company, Battle Creek, Michigan, USA) ready-to-eat cereal with 2% milk. A low fibre, RS-free breakfast was chosen as the standard breakfast meal to ensure that any effects observed were a result of prior potato RS consumption. The RS content of the test and control foods was determined from published reports [17,27,28], and other nutritional information was determined using food label information, when available, and the US Department of Agriculture's FoodData Central [29]. Summaries of the nutritional attributes of the potato and control meals and snacks, and the low-fibre, RS-free standard breakfast meal, are provided in Table 1.

Study protocol

This study was a randomised, two-period cross-over study, with at least a 7-day washout period between the test days. Both treatments consisted of the same standard low-fibre, RS-free breakfast and isocaloric lunch meals, evening meals and evening snacks that differed in RS

content (Table 1). The same low-fibre, RS-free breakfast was provided the morning following both treatments as part of a meal tolerance test (MTT).

At the first clinic visit, subjects underwent fasting capillary glucose and HbA1c testing, provided body weight, height and waist circumference measurements and medical history information. Body weight was measured on a digital scale with the subject wearing light clothing and no shoes. Eligible subjects provided seated resting heart rate and blood pressure, which was measured three times, each separated by 1 min, after the subject had been resting quietly for 5 min; the three measurements were recorded and averaged. Subjects were given a 3-day diet record and were instructed to complete it on two weekdays and one weekend day prior to each test meal visit. They were also provided a low fibre, RS-free standard breakfast (2100 kJ, 81 g of carbohydrate, 19 g of protein, 9 g of fat) with instructions to consume it on the morning of their next clinic visit. Subjects were instructed to avoid vigorous physical activity and alcohol consumption for 24 h prior to test visits and to avoid tobacco products for 1 h prior to testing and through the duration of the test procedures because nicotine-containing products have been shown to acutely impact FFA [30].

On the day of the second clinic visit (7–14 days after first visit), subjects consumed the standard breakfast meal at home and arrived at the clinic prior to lunch. Body weight, heart rate and blood pressures were assessed at the visit and every subsequent visit thereafter. Three-day diet records were collected from each subject. Each subject was then randomised, using a computer-generated randomisation scheme generated by the study statistician, to either the potato intervention or control condition for the first treatment. An envelope was opened at the time of randomisation of a subject to a diet sequence. As a result of the nature of the study treatments, neither subjects, nor staff were blinded, although the statistician was blinded to treatment during the initial analyses. Both the lunch and evening meals were consumed at the clinic. The subjects took home an evening snack meal and consumed it chilled, 2 h prior to bedtime (~12 h prior to the next day's standard breakfast meal). Subjects were instructed to not eat any additional foods, other than those provided, on the day prior to the test. Subjects subsequently filled out a palatability questionnaire after each of the meals.

The next day, after an overnight fast, subjects returned to the clinic for a MTT. They were provided the low-fibre, RS-free standard breakfast and asked to complete the meal within 20 min. Blood samples were collected for plasma insulin, glucose and FFA measurements and visual analogue scale (VAS) assessments for appetite (fullness, hunger, desire to eat, and prospective food consumption) were administered at pre-breakfast ($t = -5$) and at $t = 30$, 60, 90, 120, 150, 180, 210, 240, 270 and 300 ± 2 min, where $t = 0$ min was the start of consumption of the standard breakfast meal. Breath hydrogen levels were assessed pre-breakfast ($t = -5$) and after the blood collections at $t = 60$, 120, 180, 240 and 300 min. The subjects then completed a washout period of no less than 7 days after which they returned to the clinic to repeat the study procedures with the other condition.

Blood insulin, glucose and free fatty acids

Fasting plasma glucose was assessed using an enzymatic colorimetric assay (Cleveland HeartLab, Cleveland, OH, USA) and fasting plasma insulin was assessed using an electrochemiluminescence immunoassay (Cleveland HeartLab). Plasma FFA were analysed by OmegaQuant (Sioux Falls, SD, USA) [31].

Breath hydrogen

End-expiratory breath samples were collected using the Easy Sampler™ with tube holder (Quintron Instrument Company, Milwaukee, WI, USA). Hydrogen was measured by gas chromatography (Quintron Microlyzer, Model SC; Quintron Instrument Company Inc., Milwaukee, WI, USA) in both breath samples and simultaneously obtained room air.

Dietary composition analysis

All foods, beverages and supplements consumed over 3 days prior to test days were recorded in the 3-day diet record. The diet records were analysed using FOOD PROCESSOR® NUTRITION ANALYSIS & FITNESS SOFTWARE, version 10.4 or later (ESHA Research, Salem, OR, USA).

Visual analogue scale assessments

Subjective appetite was assessed by subjects self-reporting the level of fullness, hunger, desire to eat and prospective food consumption on a 100-mm line VAS. The scales were anchored with extremes. Hunger was anchored with 'Not hungry at all' and 'As hungry as I have ever felt'. Fullness was anchored with 'Not at all full' and 'As full as I have ever felt". Desire to eat was anchored with 'Not at all strong' and 'As strong as I've ever felt'. Although desire to eat is similar to hunger, hunger is typically perceived as a physical sensation, whereas desire to eat may be more sensitive to other psychological cues of appetite that may drive the desire to eat (e.g. boredom). Prospective consumption attempts to provide a qualitative assessment of the amount of food a person could eat by asking

'How much do you think you could eat right now?' and the question is anchored with 'Nothing at all' or 'A large amount'.

Statistical analysis

Because this was designed as a pilot study, an evaluable sample of 20 subjects was expected to provide 80% power for a one-sided alpha of 0.05 to detect a 12% difference between treatment conditions in the MISI, assuming a 20% pooled SD, and 80% power to detect differences for key secondary outcome variables with effect sizes of at least 0.6 SDs. Again, because this was a pilot study, a one-sided alpha was used to minimise risk of type 2 statistical error. To account for attrition and non-adherence to the study protocol, a sample of 24 subjects was randomised.

All statistical analyses were conducted using SPSS, version 24.0 (IBM Corp., Armonk, NY, USA). The evaluable study sample included all subjects who provided valid data during both test conditions. Comparisons between the diet condition sequence groups for baseline characteristics (such as age, sex, body weight, BMI, and waist circumference) were assessed by analysis of variance (ANOVA) and chi-squared tests, or other techniques as appropriate. All tests of significance, unless otherwise stated, were performed at alpha = 0.05 (one-sided). MISI was considered the primary outcome, and no adjustment was applied to the alpha level for tests of non-primary outcome variables.

For continuous variables, the difference in responses at the end of each test condition was assessed using repeated measures ANOVA. Initial ANOVA models contained terms for diet condition, sequence and period, with subject as a random effect. Model residuals were assessed for normality. For variables that showed evidence of non-normality based on histogram and q-q plots, values were ranked prior to running the final model. Models were reduced using backward selection until only significant ($P < 0.05$) terms or diet condition remained in the model. Separate models were run for key variables to assess possible carry-over effects (diet condition by sequence interactions). Because statistically significant or clinically important interactions were not observed, only pooled data from both treatment sequences are presented. Descriptive statistics presented include frequencies and percentages for categorical variables, mean (SEM) for normally distributed variables, and median (interquartile limits) for non-normally distributed variables.

The MISI was calculated from plasma glucose and insulin concentrations pre-MTT and during the 120 min postprandial period as described previously [32] using: $10\,000/(G_0 \times I_0 \times G_m \times I_m)^{0.5}$, where G_0 and I_0 are premeal values for glucose (G) and insulin (I), respectively, and G_m and I_m are the respective mean post-meal values during the 120 min of the test. Total area under the curve (AUC) and net incremental area under the curve (niAUC) for selected variables were calculated using the trapezoidal rule with methods outlined by Brouns *et al.* [33]. The homeostasis model assessment-2 (non-linear model) values for insulin sensitivity (HOMA2-%S) and pancreatic beta-cell function (HOMA2-%B) were calculated as described previously using an online calculator (https://www.dtu.ox.ac.uk/homacalculator/index.php) [34].

Results

Subject disposition and baseline characteristics

In total, 37 subjects were screened and 24 met entry criteria and were randomised to a diet condition. Of those randomised, five subjects dropped out of the study as a result of poor vein access at a subsequent clinic visit ($n = 3$) or failure to comply with study procedures ($n = 2$) (Fig. 1). The baseline characteristics of the evaluable subjects are shown in Table 2. There were no significant changes from baseline within the test conditions for blood pressure, body weight or heart rate (see Supporting information, Table S2). In the potato intervention, one subject reported mild gastric upset with symptoms similar to gastroesophageal reflux. It was judged this was not related to the study product and symptoms resolved duriing the same day. In the control condition, one subject reported lightheadedness and one subject reported headache, dizziness and nausea that were not related to the study product.

Insulin sensitivity and related markers

Values for variables related to CHO metabolism in the control and potato conditions are shown in Table 3. Values for MISI, HOMA2-%S and HOMA2-%B were higher during the potato intervention, although the differences did not approach statistical significance. Postprandial insulin and glucose responses to the standard breakfast meals are shown in Fig. 2. Fasting plasma glucose was significantly lower after the potato intervention ($P = 0.043$). Fasting plasma insulin was also lower after the potato intervention, although this did not reach significance ($P = 0.077$).

Breath hydrogen and plasma free fatty acids

Breath hydrogen and plasma FFA values are shown in Figs 3 and 4 and Table 4. Breath hydrogen at 300 min (end of test) was significantly higher ($P = 0.037$) and plasma FFA at 300 min was significantly lower ($P = 0.039$) in the potato condition compared to the control condition. Breath hydrogen AUC was higher during the potato condition, although this did not approach

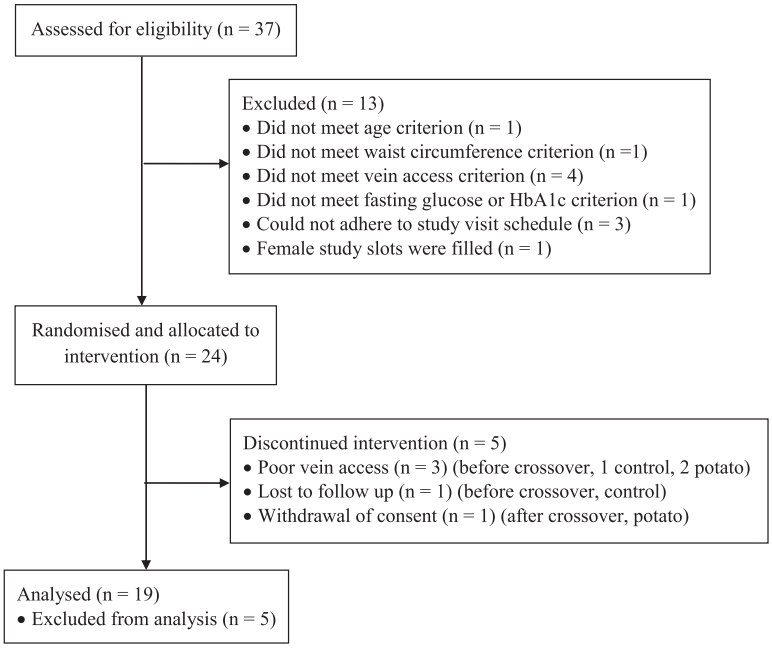

Figure 1 Flow diagram of subjects assessed for eligibility, excluded, randomised, and analysed for the present study.

Table 2 Demographic and anthropometric baseline characteristics of subjects (n = 19)

	n (%)
Gender	
Female	13 (68.4)
Male	6 (31.6)
Race	
White	11 (57.9)
Black/African American	7 (36.8)
Not disclosed	1 (5.3)
Ethnicity	
Not Hispanic/Latino	12 (63.2)
Hispanic/Latino	4 (21.1)
Not disclosed	3 (15.8)
Smoking status	
Non-smoker	16 (84.2)
Current smoker	2 (10.5)
Alcohol consumer	
No	12 (63.2)
Yes	7 (36.8)
	Mean (SEM)
Age (years)	48.0 (2.9)
Height (cm)	167 (2.3)
Weight (kg)	92.6 (3.6)
BMI (kg m^{-2})	33.1 (0.9)
Waist circumference (cm)	
Male	110.0 (3.3)
Female	107.4 (3.1)
Fasting capillary glucose (mmol L^{-1})	5.1 (0.2)

BMI, body mass index; *n*, number of subjects; SEM, standard error of the mean.

significance. However, FFA AUC was significantly lower during the potato condition (P = 0.028).

Appetite

VAS assessments of appetite (hunger, desire to eat, fullness and prospective consumption) are shown in Fig. 5 and Table 5. Hunger, desire to eat and prospective consumption niAUC values did not differ between the potato intervention and control conditions. Fullness niAUC was significantly higher in the control compared to the potato intervention (P = 0.002).

Dietary intake and palatability

Dietary intake, as measured with a 3-day diet record, was not different between the diet conditions for energy, total CHO, protein, fat, saturated fat and dietary fibre (see Supporting information, Table S1). Palatability ratings were assessed with a nine-point Likert scale ranging from 1 (dislike extremely) to 9 (like extremely) with 5 representing neither like nor dislike. Overall liking did not differ between potato and control meals (see Supporting information, Table S3).

Discussion

We hypothesised that the intake of RS-enriched potatoes, versus isocaloric CHO-based control foods, would increase insulin sensitivity as a result of the ability of RS

Table 3 Matsuda insulin sensitivity index (MISI), homeostasis model assessment-2 insulin sensitivity (HOMA2-%S) and beta-cell function (HOMA2-%B) following a breakfast meal tolerance test by diet condition

	Condition		
	Potato (n = 19)*	Control (n = 19)*	Between condition P-value
Median (Q1, Q3)			
MISI† (mg dL^{-1}*μU $L^{-1})^{-1}$	4.08 (2.70, 6.79)	3.88 (2.44, 7.50)	0.310
HOMA2-%S	94.8 (52.0, 149)	70.3 (49.8, 121)	0.094
HOMA2-%B	136 (84.8, 157)	119 (92.2, 170)	0.398
Mean (SEM)			
Fasting plasma glucose (mmol L^{-1})	4.4 (0.2)	4.8 (0.2)	**0.043**
Fasting plasma insulin (pmol L^{-1})	48.0 (34.2, 92.4)	63.6 (39.6, 96.0)	0.077

Q1 and Q3, interquartile limits.
Bolded P values are considered significant.
*For MISI n = 18 because one subject was missing glucose and insulin values at t = 60 min and was excluded from this analysis.
†MISI was calculated using glucose and insulin valued at t = 0, 30, 60, 90 and 120 min.

to enhance colonic fermentation and downstream processes. Although the insulin sensitivity results of our study were equivocal, the increase in late postprandial breath hydrogen, an indicator of colonic fermentation, and the decrease in late postprandial plasma FFA are consistent with the mechanistic hypothesis. Furthermore, fasting plasma glucose was significantly lower after the potato intervention compared to the control and fasting plasma insulin showed a trend toward being lower. These findings are therefore suggestive of effects consistent with those hypothesised, although the sample size was perhaps too small to show statistically significant differences for insulin sensitivity indices. Although the 5% difference between conditions for MISI did not approach statistical significance, that for HOMA2-%S showed a one-sided P-value of 0.09 with a value that was 34.9% higher during the potato condition. A post-hoc sample size calculation was completed using the observed variability in the log-transformed values for these variables to account for non-normality in the distributions. With the magnitudes of the differences observed in the present study, assuming the same variability, an evaluable sample of 28 subjects would have provided 80% power to detect these differences, with two-sided alpha levels of 0.05. Therefore, a study in a larger sample may achieve the power needed to detect significant and clinically important differences.

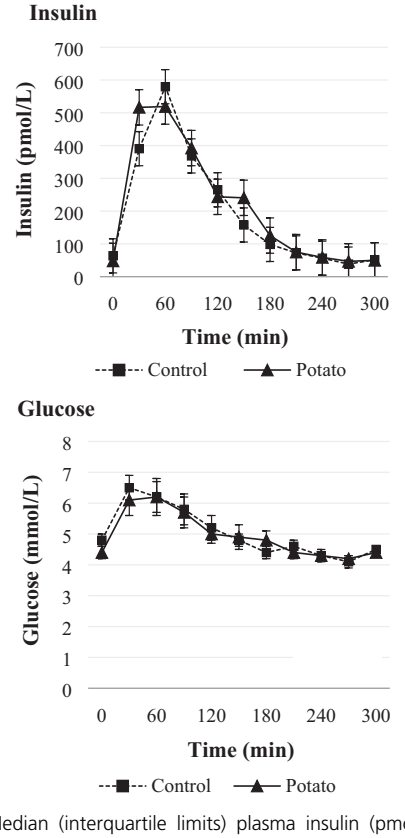

Figure 2 Median (interquartile limits) plasma insulin (pmol L^{-1}) and mean (SEM) glucose (mmol L^{-1}) concentrations from pre-breakfast (t = 0 min) to 300 min post-breakfast by diet condition (n = 19).

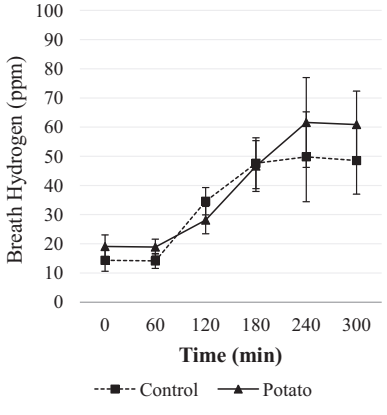

Figure 3 Mean (SEM) breath hydrogen (ppm) from pre-breakfast (t = 0 min) to 300 min post-breakfast by diet condition (n = 17).

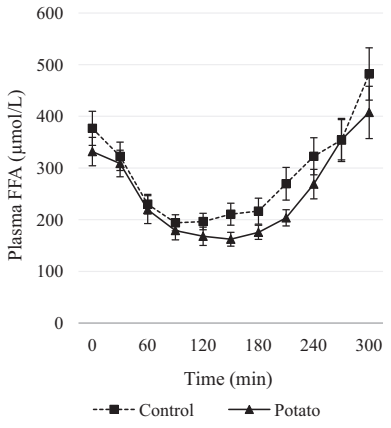

Figure 4 Mean (SEM) plasma free fatty acid concentration (μmol L^{-1}) from pre-breakfast (t = 0 min) to 300 min post-breakfast by diet condition (n = 19).

The consumption of various types of RS has been reported to produce favourable effects on several aspects of health, including markers of glucose and lipid metabolism [3,6,18,19,21,22,35-37]; although, not all studies have shown a benefit [4,38]. Interestingly, only one of these studies [21] has investigated RS type 3 present in potatoes, although it did not include measures of insulin sensitivity. Prior research by our group has shown that RS intake can improve markers of metabolic health [3,19,22]; however, gender and baseline insulin sensitivity may play a role in the effects of RS on insulin sensitivity. In one study, we showed that 4 weeks of supplemental RS type 2 intakes of both 15 g day^{-1} and 30 g day^{-1} increased insulin sensitivity in overweight and obese men, but not women, compared to 0 g day^{-1} of supplemental intake [19]. Importantly, the sample in the present study was predominantly (68%) women, which may partially explain the lack of effect on insulin sensitivity. It is unclear why the effect of RS on insulin sensitivity would be different for women. Hoeg *et al.* [39] showed that increasing FFA concentrations via infusion of a lipid emulsion decreased insulin sensitivity in men, but not women, suggesting that women may be less sensitive to changes in FFA. Some studies have shown improvements in insulin sensitivity and glycaemia in individuals with insulin resistance or impaired glucose tolerance at baseline [18,21]. Patterson *et al.* [21] recently found lower early postprandial (15–30 min) glucose and insulin levels in women with elevated fasting glucose and insulin after consuming approximately 10–11 g of RS from cooked and cooled Russet potatoes compared to boiled, non-chilled potatoes. Gower *et al.* [18] also found that women with insulin resistance at baseline showed increased insulin sensitivity after the intake of 30 g day^{-1} of RS type 2, although no effect was observed in women who were more insulin sensitive (top 39% of the insulin sensitivity index distribution in the study sample). Because this was a pilot study, we did not assess baseline insulin sensitivity, although a median HOMA2-%S value of 70.3 during the control condition (where a value of 100 reflects the level of insulin sensitivity in lean, 40-year-old men) [34] suggests that many of the participants in the present study likely had some degree of insulin resistance. Therefore, subsequent studies on RS-enriched potatoes may need to consider gender and baseline insulin sensitivity when determining effects on markers of glucose and insulin metabolism.

A previous study conducted by our lab measured insulin sensitivity after 4 weeks of supplemental RS type 2 [19], although several of the metabolic effects of RS intake can be measured after a short period of consumption [6,20-22] and, for this reason, a 24-h intervention was selected for the present pilot study. Robertson *et al.* [6] reported that intake of a low-residue diet, supplemented with 60 g RS type 2 over a 24-h period, compared to the same diet without supplementation, lowered postprandial plasma glucose and insulin and increased insulin sensitivity and breath hydrogen output during the 5 h following the

Table 4 Breath hydrogen and free fatty acid responses during a breakfast meal tolerance test by diet condition

	Condition		
	Potato (n = 19)	Control (n = 19)	Between condition P-value
Mean (SEM)			
Breath hydrogen†, $AUC_{0–300\ min}$ (ppm*min)	11 756 (1806)	10 749 (1298)	0.280
Free fatty acids, $AUC_{0–300\ min}$ (μmol L^{-1}*min)	72 457 (5932)	82 722 (6848)	**0.028**
Free fatty acids at 300 min (μmol L^{-1})	408 (50.6)	482 (50.6)	**0.039**
Median (Q1, Q3)			
Breath hydrogen at 300 min (ppm)	53 (22, 96)	43 (22, 62)	**0.037**

$AUC_{0–300\ min}$, area under the curve from 0-300 min; ppm, parts per million; Q1 and Q3, interquartile limits.
Bolded P values are considered significant.
†Two subjects did not have baseline values and were excluded from the analysis (n = 17).

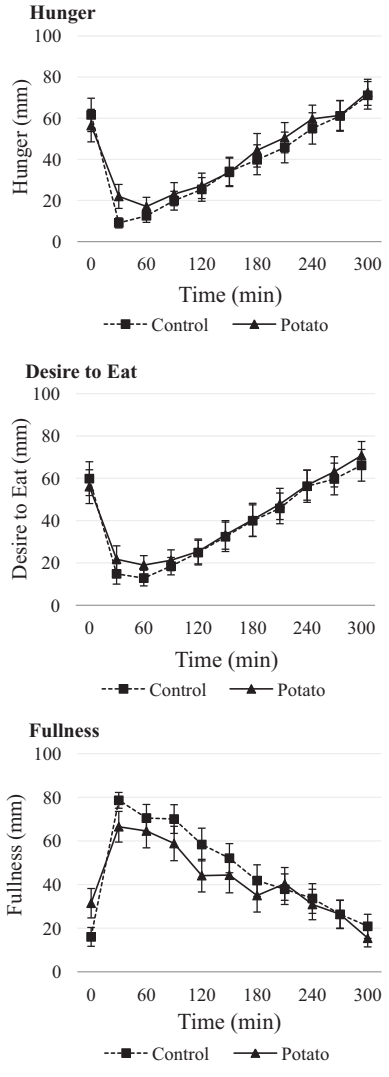

Figure 5 Mean (SEM) visual analogue scale ratings from pre-breakfast (t = 0 min) to 300 min post-breakfast for hunger, desire to eat, and fullness by diet condition (n = 19). Only fullness had a significantly different net incremental area under the curve (niAUC) between control and potato (P = 0.003).

24-h treatment period [6]. Additionally, FFA levels were suppressed in the late postprandial period after the breakfast meal in the same study [6]. Similarly, a single bolus intake of 28 g of RS and 75 g of glucose has been shown to significantly reduce the postprandial serum glucose, insulin and FFA responses compared to an intake of 75 g of glucose alone [20]. In that study, postprandial breath hydrogen levels were significantly increased following the RS compared to the intake of glucose, indicating increased RS-mediated colonic fermentation [20]. In the present study, a more practical level of intake for RS (~18 g) was evaluated. Although differences were observed in fasting plasma glucose and FFA, insulin sensitivity did not differ significantly between conditions, possibly as a result of the small sample size.

The quantity of potatoes consumed in the present study (300 g containing ~18 g RS) was chosen because it represents a level of intake (approximately two potatoes) that can be reasonably achieved. Several other studies [6,20] have demonstrated significant effects on insulin, glucose, breath hydrogen and FFA, although, at higher doses, this may prove difficult for many individuals to achieve. The study by Robertson *et al.* [6] demonstrated changes in insulin and glucose metabolism, and breath hydrogen with the consumption of 60 g RS within 24 h. Similarly, the study by Rahat-Rozenbloom *et al.* [20] showed a benefit on postprandial serum glucose, insulin and FFA responses with a single bolus of 28 g of RS. The present study showed similar metabolic results with only 18 g of RS; however, a larger study is still needed to confirm these findings and to determine whether these metabolic changes may contribute to improved insulin sensitivity.

In the present pilot study, no significant differences were observed for hunger or desire to eat VAS ratings but, surprisingly, fullness ratings were significantly lower postprandially after the RS intake (P = 0.003). It has been hypothesised that the effects of RS fermentation may be able to impact appetite at subsequent meals [36,40] and thus appetite was assessed >12 h after consuming RS. Mixed results have been reported in studies with respect to the acute and chronic effects of consuming RS on appetite ratings and food intake [21,22,41]. Similar to the present study, Patterson *et al.* [21] found an increase in subjective hunger 15–60 min after consuming 200 g of cooked then chilled potatoes compared to boiled potatoes that were not chilled. This was attributed to differences in palatability between the chilled potato treatment and the boiled potato control; however, the present study used a chilled egg noodle control and did not show any significant differences in palatability between the treatment and control. Alternatively, our group has previously shown that consumption of a breakfast scone containing 16.5 g of type 4 RS significantly reduced hunger and desire to eat compared to a control scone, even though the control scone contained higher levels of available CHO and energy [22]. Both of these studies [21,22] measured appetite immediately after consuming RS, whereas the present study measured appetite >12 h after consuming RS and following a low-fibre, low-RS meal. Additional investigation is needed to further assess the effects of RS consumption on appetite, both acutely and at subsequent meals. Moreover, longer studies will be needed to assess whether potential effects on appetite persist with chronic consumption. Furthermore, the RS content of the meals in the present study, although practical, may be too low to impact appetite over the long term.

Table 5 Net incremental area under the curve (niAUC) for hunger, desire to eat, fullness and prospective consumption visual analogue scale (VAS) ratings from pre-breakfast (t = 0 min) to 300 min post-breakfast

	Condition		
	Potato (n = 19)	Control (n = 19)	Between condition P-value
Mean (SEM)			
Hunger (mm*min)	–4825 (1787)	–7417 (2367)	0.150
Desire to eat (mm*min)	–5048 (1687)	–6910 (2221)	0.221
Fullness (mm*min)	3591 (1567)	9819 (1726)	**0.002**
Prospective consumption (mm*min)[†]	–4479 (1871)	–8043 (1536)	0.064

Bolded P values are considered significant.

[†]One subject was missing t = 30-minute VAS rating during the control breakfast and was excluded from the analysis (n = 18).

The strengths of the present study include the use of commonly consumed foods for both diet conditions, in realistic quantities (approximately two potatoes over 24 h or isocaloric, CHO-based foods) and the incorporation of these foods into mixed meals. Potatoes are versatile and a commonly consumed food that can be readily incorporated into the habitual diet as a source of RS. Future, longer-term studies may utilise potatoes in conjunction with other sources of RS (e.g. certain grains) to allow greater variety. The present pilot study was limited by a small sample size. A larger sample may provide adequate statistical power to detect significant differences for insulin sensitivity measures, which may be clinically relevant. The sample size calculations completed during the planning stage were based on a difference of 12% in MISI, which is larger than the observed difference of 5%. HOMA2-%S showed a larger difference, with a value that was 35% higher during the potato intervention compared to the control. However, because this is a less precise measure of insulin sensitivity, the sample size was not sufficiently large to demonstrate statistical significance for this difference [(34)]. If confirmed in a larger sample, this would likely be a clinically important difference [(42)]. Another limitation is the use of a one-tailed test, which increases the likelihood of a Type 1 statistical error. A one-tailed test was used because this was a pilot study with a small sample size with the objective of determining whether a full-scale study was justified. An additional limitation of this pilot study was the lack of compositional testing of the foods to determine the precise RS content instead of relying on published data. Although it is not uncommon to rely on published data [(7,21)] or data from manufacturers, [(6,19,35)] a future, full-scale study should include compositional testing to determine accurate RS content. Finally, a full-scale study should include baseline measures of insulin sensitivity during habitual diet consumption.

In summary, the results of this pilot study showed significantly lower fasting glucose, increased breath hydrogen and reduced FFA concentrations in the late postprandial period after the intake of RS-enriched potatoes versus control foods. There was also a trend toward lower fasting insulin. However, differences in MISI and other markers of insulin sensitivity did not reach statistical significance. Additional research in a larger study sample is warranted to further assess the potential for consumption of potato RS to favourably alter glucose metabolism. Further studies may also benefit from evaluating multiple sources of RS from commonly consumed food, including potatoes, to evaluate the long-term feasibility of integrating more RS into the diet.

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Conflict of interests, funding and authorship

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OMP, MRD, CEM and KCM contributed to the design of the study. CEM conducted the study (including recruitment, preparation of test foods and sample collection). MW statistically analysed the data. OMP and LMS drafted the manuscript; All authors contributed to the interpretation of the study and have read and approved the final version submitted for publication.

Transparency declaration

The lead author affirms that this manuscript is an honest, accurate and transparent account of the study being reported. The reporting of this work is compliant with CONSORT guidelines. The lead author affirms that no important aspects of the study have been omitted and that any discrepancies from the study as planned (ClinicalTrials.gov: NCT03689738) have been explained.

References

1. Saeedi P, Petersohn I, Salpea P *et al.* (2019) Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: Results from the International Diabetes Federation Diabetes Atlas, 9th edition. *Diab Res Clin Pract* **157**, 107843.
2. Wölnerhanssen BK, Cajacob L, Keller N *et al.* (2016) Gut hormone secretion, gastric emptying, and glycemic responses to erythritol and xylitol in lean and obese subjects. *Am J Physiol Endocrinol Metab* **310**, E1053–E1061.
3. Maki KC & Phillips AK (2015) Dietary substitutions for refined carbohydrate that show promise for reducing risk of type 2 diabetes in men and women. *J Nutr* **145**, 159S–163S.
4. Peterson CM, Beyl RA, Marlatt KL *et al.* (2018) Effect of 12 wk of resistant starch supplementation on cardiometabolic risk factors in adults with prediabetes: a randomized controlled trial. *Am J Clin Nutr* **108**, 492–501.
5. Robertson MD, Bickerton AS, Dennis AL *et al.* (2005) Insulin-sensitizing effects of dietary resistant starch and effects on skeletal muscle and adipose tissue metabolism. *Am J Clin Nutr* **82**, 559–567.
6. Robertson MD, Currie JM, Morgan LM *et al.* (2003) Prior short-term consumption of resistant starch enhances postprandial insulin sensitivity in healthy subjects. *Diabetologia* **46**, 659–665.
7. Dodevska MS, Sobajic SS, Djordjevic PB *et al.* (2016) Effects of total fibre or resistant starch-rich diets within lifestyle intervention in obese prediabetic adults. *Eur J Nutr* **55**, 127–137.
8. Diabetes Prevention Program Research Group (2005) Role of Insulin Secretion and Sensitivity in the Evolution of Type 2 Diabetes in the Diabetes Prevention Program. *Diabetes* **54**, 2404–2414.
9. Asp NG, Bjorck I, Holm J *et al.* (1987) Enzyme resistant starch fractions and dietary fibre. *Scand J Gastroenterol* **12** (Suppl 129), 29–32.
10. Birt DF, Boylston T, Hendrich S *et al.* (2013) Resistant starch: promise for improving human health. *Adv Nutr* **4**, 587–601.
11. Keenan MJ, Zhou J, Hegsted M *et al.* (2015) Role of resistant starch in improving gut health, adiposity, and insulin resistance. *Adv Nutr* **6**, 198–205.
12. Kimura I, Inoue D, Hirano K *et al.* (2014) The SCFA receptor GPR43 and energy metabolism. *Front Endocrinol* **5**, https://doi.org/10.3389/fendo.2014.00085.
13. Boden G (1997) Role of fatty acids in the pathogenesis of insulin resistance and NIDDM. *Diabetes* **46**, 3–10.
14. Bajaj M, Suraamornkul S, Romanelli A *et al.* (2005) Effect of a sustained reduction in plasma free fatty acid concentration on intramuscular long-chain fatty Acyl-CoAs and insulin action in type 2 diabetic patients. *Diabetes* **54**, 3148–3153.
15. Oh YS, Bae GD, Baek DJ *et al.* (2018) Fatty acid-induced lipotoxicity in pancreatic beta-cells during development of type 2 diabetes. *Front Endocrinol* **9**, 384.
16. Englyst KN, Liu S & Englyst HN (2007) Nutritional characterization and measurement of dietary carbohydrates. *Eur J Clin Nutr* **61**, S19–S39.
17. Raatz SK, Idso L, Johnson LK *et al.* (2016) Resistant starch analysis of commonly consumed potatoes: Content varies by cooking method and service temperature but not by variety. *Food Chem* **208**, 297–300.
18. Gower BA, Bergman R, Stefanovski D *et al.* (2016) Baseline insulin sensitivity affects response to high-amylose maize resistant starch in women: a randomized, controlled trial. *Nutr Metab* **13**, 2.
19. Maki KC, Pelkman CL, Finocchiaro ET *et al.* (2012) Resistant starch from high-amylose maize increases insulin sensitivity in overweight and obese men. *J Nutr* **142**, 717–723.
20. Rahat-Rozenbloom S, Fernandes J, Cheng J *et al.* (2017) The acute effects of inulin and resistant starch on postprandial serum short-chain fatty acids and second-meal glycemic response in lean and overweight humans. *Eur J Clin Nutr* **71**, 227–233.
21. Patterson MA, Fong JN, Maiya M *et al.* (2019) Chilled potatoes decrease postprandial glucose, insulin, and glucose-dependent insulinotropic peptide compared to boiled potatoes in females with elevated fasting glucose and insulin. *Nutrients* **11**, 2066.
22. Stewart ML, Wilcox ML, Bell M *et al.* (2018) Type-4 resistant starch in substitution for available carbohydrate reduces postprandial glycemic response and hunger in acute, randomized, double-blind controlled study. *Nutrients* **10**, 129.
23. National Heart Lung Blood Institute, National Institute of Diabetes Digestive Kidney Diseases, Expert Panel on the Identification and Treatment of Overweight Obesity in Adults (1998) *Clinical Guidelines on the Identification, Evaluation, and Treatment of Overweight and Obesity in Adults: The Evidence Report*. National Institutes of Health, National Heart, Lung, and Blood Institute.
24. World Medical Association (2000) World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. *JAMA* **284**, 3043–3045.

25. Muir JG & O'Dea K (1992) Measurement of resistant starch: factors affecting the amount of starch escaping digestion in vitro. *Am J Clin Nutr* **56**, 123–127.
26. Dupuis JH, Liu Q & Yada RY (2014) Methodologies for increasing the resistant starch content of food starches: A review. *Comp Rev Food Sci Food Safe* **13**, 1219–1234.
27. Murphy MM, Douglass JS & Birkett A (2008) Resistant starch intakes in the United States. *J Am Diet Assoc* **108**, 67–78.
28. Gelroth JA & Ranhotra GS (2000) Determination of resistant starch in selected grain-based foods. *J AOAC Int* **83**, 988–991.
29. US Department of Agriculture ARS (2019) Food Data Central. Available at: fdc.nal.usda.gov
30. Bajaj M (2012) Nicotine and insulin resistance: when the smoke clears. *Diabetes* **61**, 3078–3080.
31. Miles JM, Wooldridge D, Grellner WJ *et al.* (2003) Nocturnal and postprandial free fatty acid kinetics in normal and type 2 diabetic subjects: effects of insulin sensitization therapy. *Diabetes* **52**, 675–681.
32. Maki KC, Kelley KM, Lawless AL *et al.* (2011) Validation of insulin sensitivity and secretion indices derived from the liquid meal tolerance test. *Diab Technol Therap* **13**, 661–666.
33. Brouns F, Bjorck I, Frayn KN *et al.* (2005) Glycaemic index methodology. *Nutr Res Rev* **18**, 145–171.
34. Wallace TM, Levy JC & Matthews DR (2004) Use and abuse of HOMA modeling. *Diabetes Care* **27**, 1487–1495.
35. Maki KC, Sanders LM, Reeves MS *et al.* (2009) Beneficial effects of resistant starch on laxation in healthy adults. *Int J Food Sci Nutr* **60**(Suppl 4), 296–305.
36. Zhang L, Li HT, Shen L *et al.* (2015) Effect of dietary resistant starch on prevention and treatment of obesity-related diseases and its possible mechanisms. *Biomed Environ Sci* **28**, 291–297.
37. Lockyer S & Nugent AP (2017) Health effects of resistant starch. *Nutr Bull* **42**, 10–41.
38. Marlatt KL, White UA, Beyl RA *et al.* (2018) Role of resistant starch on diabetes risk factors in people with prediabetes: design, conduct, and baseline results of the STARCH trial. *Contemp Clin Trials* **65**, 99–108.
39. Høeg LD, Sjøberg KA, Jeppesen J *et al.* (2011) Lipid-induced insulin resistance affects women less than men and is not accompanied by inflammation or impaired proximal insulin signaling. *Diabetes* **60**, 64–73.
40. Ibrugger S, Vigsnaes LK, Blennow A *et al.* (2014) Second meal effect on appetite and fermentation of wholegrain rye foods. *Appetite* **80**, 248–256.
41. Al-Mana NM & Robertson MD (2018) Acute effect of resistant starch on food intake, appetite and satiety in overweight/obese males. *Nutrients* **10**, 1993.
42. Faerch K, Vaag A, Holst JJ *et al.* (2009) Natural history of insulin sensitivity and insulin secretion in the progression from normal glucose tolerance to impaired fasting glycemia and impaired glucose tolerance: the Inter99 study. *Diabetes Care* **32**, 439–444.

Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1. Dietary intake of subjects prior to potato and control conditions based on 3-day diet records.

Table S2. Change from baseline to end of treatment in blood pressure, body weight and heart rate by test condition.

Table S3. Overall liking of study product and control meals.